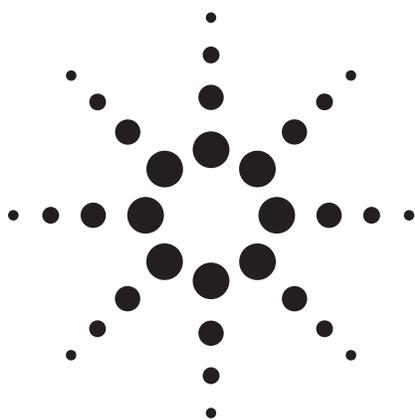


Multi-Element Determination of Heavy Metals in Dietary Supplements Using Collision/Reaction Cell ICP-MS



Application

Foods

Authors

Emma Peachey, Ruth Hearn, and Selvarani Elahi
LGC, Queens Road
Teddington, Middlesex, TW11 0LY
UK

Abstract

Determination of 11 metals (including arsenic, chromium, mercury, iron, copper, nickel, zinc, selenium, lead, cadmium, and thallium) in UK-consumed dietary supplements was carried out using ICP-MS. The instrument used was the Agilent 7500ce, which is equipped with a collision/reaction cell (Octopole Reaction System), and was operated in no-gas, helium, and hydrogen modes, all acquired within a single method. Samples were microwave digested with nitric acid/hydrogen peroxide/hydrofluoric acid and quantified using external calibration. The method was assessed by the analysis of two certified reference materials (LGC7160 and SRM1577b), and recoveries were $100 \pm 15\%$ of the certified value for all elements.

Introduction

Heavy metals are natural components of the earth's crust and are widely used in agricultural, construction, manufacturing, and food/material processing industries. As trace elements, some heavy metals are metabolically essential to humans at low levels, but at higher concentrations they can become toxic. Heavy metal toxicity can result from high ambient air concentrations near emission sources, drinking-water contamination, or intake via the food chain.

Conversely, as our lifestyles have become increasingly busy, leaving less time to prepare and eat

well-balanced meals, our diets are at an increased risk of becoming nutrient deficient. Dietary supplements, sold in capsule, tablet, or liquid form offer a simple and convenient way to supplement our diets and reduce the risk of nutrient deficiency.

In the UK, most products described as dietary supplements are regulated as foods and are subject to the general provisions of The Food Safety Act 1990 and secondary legislation on safety and labeling (Food Labeling Regulations 1996 [as amended] and the Food Supplements Directive 2002/46/EC). Past investigations have shown that some dietary supplements can contain elevated concentrations of metals and other elements. While some metals, such as selenium, iron, copper, chromium, and zinc [1], are essential at low concentrations, others, such as arsenic, cadmium, lead, and mercury, are toxic [2]. In the UK, arsenic is the only element for which there is legislation on permitted levels in foods. Arsenic is regulated under the "Arsenic in Food (as amended) Regulations 1959," which states a limit of 1 mg/kg in "general" food [3]. Cadmium, lead, mercury, and tin in specific foods are regulated under Commission Regulation 1881/2006 [4].

It is important that up-to-date information on the levels of metals and other elements in dietary supplements consumed in the UK is obtained, in order to assess whether there is any risk to consumers. The purpose of this study was to provide the Food Standards Agency (FSA) with up-to-date and accurate information on the concentration of a suite of metals contained within dietary supplements consumed in the UK.



To do this, microwave acid digestion was used for sample preparation, followed by multi-element determination by collision/reaction cell inductively coupled plasma mass spectrometer (CRC-ICP-MS) using He and H₂ collision gases to remove spectral interferences. The suite of metals included arsenic (As), chromium (Cr), iron (Fe), copper (Cu), nickel (Ni), zinc (Zn), selenium (Se), lead (Pb), mercury (Hg), cadmium (Cd), and thallium (Tl). Most elements were measured in standard mode (no gas) since multi-isotope ICP-MS data obtained for the food samples suggested the absence of significant polyatomic interferences. However, the ICP-MS detection of three of these elements, two essential (Fe and Se) and one toxic (As), was found to be strongly hampered by polyatomic ions. The purpose of this application is to demonstrate the ability of the Agilent 7500ce using the Octopole Reaction System (ORS) to eliminate these interferences (Table 1), providing accurate determination of these three elements in food supplements.

Experimental

Samples

Two hundred different dietary supplements (either tablet, capsule, liquid, or powder form) commercially available in the UK were sourced. The average weight of each tablet/capsule was determined using an electronic balance.

Sample Preparation

Tablets were crushed with a pestle and mortar. Crushed tablets, liquids, and powders were sub-sampled after thorough mixing. Oil capsules were digested whole. Approximately 0.7 g of sample was accurately weighed and microwave digested with 7 + 3 + 0.2 mL of nitric acid + hydrogen peroxide + hydrofluoric acid, respectively. The microwave program consisted of heating the samples to 180 °C over 20 min and holding for a further 10 min. Once cool, the digests were made up to 100 g using deionized water, and the resultant solutions were subjected to element determination by ICP-MS. Approximately 10% of the samples were digested in duplicate. A blank and QC material were included in each digestion run, containing a maximum of 12 samples.

QC Materials

Two certified reference materials were analyzed to assess the accuracy of the methodology. These were bovine liver SRM 1577b (NIST, Gaithersburg,

USA) containing 0.73 ± 0.06 mg/kg Se and 184 ± 15 mg/kg Fe and crab paste LGC7160 (LGC, Teddington, UK) containing 11 ± 1 mg/kg As.

Instrumentation

Sample digestion was undertaken in a Mars 5 microwave (CEM, Buckingham, UK). Elemental measurements were performed using an Agilent 7500ce CRC-ICP-MS operating in hydrogen mode for Se and Fe, and helium mode for As to remove spectral interferences (Table 1). All other elements were measured in standard (no gas) mode within the same method. Typical operating conditions are illustrated in Table 1. The Integrated Sample Introduction System (ISIS) was used with a pump speed set at 0.1 rps during the analysis and washout in order to minimize the amount of matrix onto the interface and optimize sample throughput.

Table 1. Instrumental Conditions for 7500ce ORS Collision/Reaction Cell Mode

Parameter	ORS Cell Mode	
	He	H ₂
Elements measured	As	Se, Fe
Spectral interferences removed by ORS gas	⁴⁰ Ar ³⁵ Cl ⁺ on ⁷⁵ As ⁺ ⁴⁰ Ca ³⁵ Cl ⁺ on ⁷⁵ As ⁺	³⁸ Ar ⁴⁰ Ar ⁺ on ⁷⁸ Se ⁺ ⁴⁰ Ar ³⁷ Cl ⁺ on ⁷⁷ Se ⁺ ⁴⁰ Ca ³⁷ Cl ⁺ on ⁷⁷ Se ⁺ ⁴⁰ Ar ¹⁶ O ⁺ on ⁵⁶ Fe ⁺ ⁴⁰ Ca ¹⁶ O ⁺ on ⁵⁶ Fe ⁺
RF power (W)	1520	
Carrier gas (L/min)	0.9	
Make-up gas (L/min)	0.26	
Nebulizer	Glass concentric, MicroMist	
Spray chamber	Quartz cooled to 2 °C	
Interface cones	Ni	
Cell gas	He	H ₂
Cell gas flow rate (mL/min)	2.5	2.2
Points per peak	3	
Repetitions	10	
Integration time per mass (sec)	0.3	

Measurement

Five-point external calibrations with standards traceable to the National Institute of Standards and Technology (NIST, Gaithersburg, USA) were used to quantify the elements in the digests. Rhodium (Rh) was used as an internal standard and added on-line (1:1 with samples). The internal standard solution also contained 4% propanol to compensate for enhancement of As and Se signal from any residual carbon in the samples. Water

Research Council's (WRC) Aquacheck solutions (used in a proficiency testing scheme) with known concentrations of the analytes (660.3 ng/g Fe, 12.6 ng/g As, and 13.27 ng/g Se) were also analyzed as independent checks on the accuracy and precision of each ICP-MS run.

Results and Discussion

All sample results were checked against legislative limits and/or limits agreed with the FSA. The action limits used were 1 mg/kg arsenic, lead, and cadmium and 0.5 mg/kg for mercury. Samples that exceeded these limits were reanalyzed to confirm the results.

As illustrated in Table 2, five samples were confirmed to contain As concentrations greater than the 1 mg/kg limit recommended in The Arsenic in Food (as amended) Regulations 1959 [3]. A further two samples were found to contain As concentrations between 0.75 mg/kg and 1 mg/kg. It should be noted that several of these supplements were derived from marine animals, and the form of the As in such materials is likely to be nontoxic arsenobetaine. The majority of supplements (> 75%) were found to contain < 0.1 mg/kg As and Se, and > 20 mg/kg Fe.

Table 2. Samples with Arsenic Concentrations Above Recommended Limit

Product	Form	As in sample (mg/kg)
Product 1	Tablet	3.3 ± 0.7
Product 2	Capsule	2.5 ± 0.6
Product 3	Capsule	20.5 ± 4.8
Product 4	Capsule	1.5 ± 0.4
Product 5	Capsule	7.3 ± 1.7

Legislative limit = 1 mg/kg [3]

The uncertainty quoted is the expanded uncertainty calculated using a coverage factor of 2, which gives a level of confidence of approximately 95%. The uncertainty was calculated based upon the principles of the Eurachem Guide [5].

None of the samples was found to contain an elevated concentration of Cd, and only one sample was close to the limit for Hg. Ten samples were confirmed to contain concentrations of Pb above 1 mg/kg.

Results in mg/kg were calculated back to mg/tablet in order to allow comparison with label claims. For a number of supplements, differences were found between the measured concentrations of Se, Fe, Zn, Cu, Cr, or Ni and the values stated on the label.

This makes it difficult for the FSA to accurately assess the dietary intake of these elements from such supplements based on label claim alone and demonstrates the necessity of this survey.

The limits of detection (LOD) and quantification (LOQ) of the described procedure, calculated according to International Union of Pure and Applied Chemistry (IUPAC) guidelines [6], are presented in Table 3. For 25% of the dietary supplements tested the Se concentration was found to be less than the LOD.

Table 3. Limits of Detection and Quantification of Se, Fe, and As

	Concentration (mg/kg) ¹		
	Se	Fe	As
LOD	0.009	0.072	0.006
LOQ	0.029	0.240	0.022

¹ Values shown are based on the average weight of dietary supplement tablet/liquid/capsule digested (0.67 g).

Recovery results for the QC materials and Aquacheck solutions were very good, with results for all elements falling within 100 ± 15% (n = > 9) of the certified/expected value. The recoveries obtained for the reference materials analyzed are illustrated in Figure 1. A number of sample and blank solutions were also spiked with Se, Fe, and As prior to microwave digestion. The resultant recoveries fell within 100 ± 10% (n = > 5) of the expected value. As a check on the repeatability of

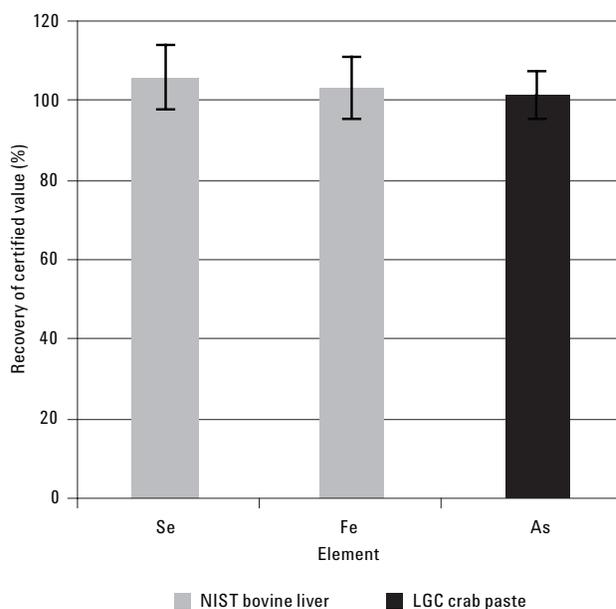


Figure 1. Recovery of Se, Fe, and As from the CRMs analyzed.

the method, approximately 10% of the samples were also independently digested and analyzed in duplicate. For these, coefficient of variation values of < 5% were found for all three elements measured in collision/reaction mode on the 7500ce ICP-MS.

Conclusions

Microwave digestion followed by analysis by ICP-ORS-MS has been shown to be a simple, reliable method for the multi-element determination of trace metals in nutritional supplements and foodstuffs. A number of supplements were found to contain Se, Fe, Zn, Cu, Cr, and Ni at concentrations that deviated from the label claim. Five samples for As and 10 samples for Pb were found to contain elevated concentrations above the recommended 1 mg/kg limit. The data generated in this survey has provided the FSA with up-to-date concentrations of these metals in a range of dietary supplements. The results have enabled the risk of metal toxicity from the consumption of dietary supplements to be assessed and published in a Food Standard Agency's Food Surveillance Information sheet [7].

References

1. World Health Organization, Trace Elements in Human Nutrition and Health, Geneva, 1996
2. The Ministry of Agriculture, Fisheries and Food (1982), Survey of Arsenic in Food. Food Surveillance Paper No. 8, Published by HMSO

3. The Arsenic in Food Regulations 1959 (S.I. [1959] No. 831), as amended by The Arsenic in Food (Amendment) Regulations 1960 (S.I. [1960] No. 2261) and The Arsenic in Food (Amendment) Regulations 1973 (S.I. [1973] No. 1052). The Stationery Office.
4. Commission Regulation (EC) No 1881/2006 of 19 December 2006 setting maximum levels for certain contaminants in foodstuffs.
5. Eurachem, Quantifying uncertainty in analytical measurement. Laboratory of the Government Chemist. London 1995 (ISBN 0 948926-08).
6. "A Statistical Overview of Standard (IUPAC and ACS) and New Procedures for Determining the Limits of Detection and Quantification: Application to Voltammetric and Stripping Techniques," *Pure & Appl. Chem.* Vol 69, No. 2, pp 297-328, 1998
7. Food Standard Agency's Food Surveillance Information sheet No 85/05 December 2005, Survey of Metals and Other Elements in Dietary Supplements.

Acknowledgements

The authors would like to thank Malcolm Burn and Kam Lee from the Food Chemistry team at LGC for performing the sample preparation/digestion, and Sheila Merson, Linda Evans, and Dave Curtis from the Specialised Techniques team for conducting the elemental determinations.

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Printed in the USA
March 19, 2008
5989-7959EN

